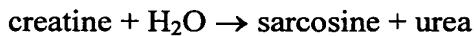


25. (Amended) A creatine amidinohydrolase (i) encoded by a nucleic acid sequence obtained by mutation of (a) the nucleic acid sequence of SEQ ID NO:2 or (b) a nucleic acid sequence encoding the amino acid sequence of SEQ ID NO:1 and (ii) having the following physicochemical properties:

Action: catalyzing the following reaction:



pH stability: being stable at pH 5-8 (40 °C, 18 h preservation)

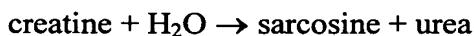
Km values for creatine in a coupling assay using a sarcosine oxidase and a peroxidase:

3.5-10.0 mM.

a¹
Cont

33. (Amended) A creatine amidinohydrolase (i) encoded by a nucleic acid sequence obtained by mutation of (a) the nucleic acid sequence of SEQ ID NO:2 or (b) a nucleic acid sequence encoding the amino acid sequence of SEQ ID NO:1 and (ii) having the following physicochemical properties:

Action: catalyzing the following reaction:



Km values for creatine in a coupling assay using a sarcosine oxidase and a peroxidase: 3.5-10.0 mM

Optimum temperature: about 40-50 °C (at a pH of about 6-8)

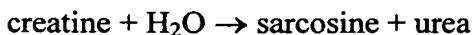
Optimum pH: pH about 8.0-9.0 (at a temperature of about 37° C)

Molecular weight: about 43,000 (SDS-PAGE).

a²

35. (Amended) A creatine amidinohydrolase (i) encoded by a nucleic acid sequence obtained by mutation of (a) the nucleic acid sequence of SEQ ID NO:2 or (b) a nucleic acid sequence encoding the amino acid sequence of SEQ ID NO:1 and (ii) having the following physicochemical properties:

Action:catalyzing the following reaction:



Heat stability: not more than about 50 °C (pH 7.5, 30 min)

pH stability: being stable at pH 5-8 (40 °C, 18 h preservation)

Km values for creatine in a coupling assay using a sarcosine oxidase and a

peroxidase: 4.5 ± 1.0 mM.

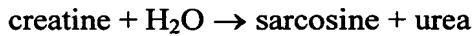
Optimum temperature: about 40-50 °C (at a pH of about 6-8)

Optimum pH: pH about 8.0-9.0 (at a temperature of about 37° C)

Molecular weight: about 43,000 (SDS-PAGE).

36. (Amended) A creatine amidinohydrolase (i) encoded by a nucleic acid sequence obtained by mutation of (a) the nucleic acid sequence of SEQ ID NO:2 or (b) a nucleic acid sequence encoding the amino acid sequence of SEQ ID NO:1 and (ii) having the following physicochemical properties:

Action: catalyzing the following reaction:



Heat stability: not more than about 50 °C (pH 7.5, 30 min)

pH stability: being stable at pH 5-8 (40 °C, 18 h preservation).

K_m values for creatine in a coupling assay using a sarcosine oxidase and a peroxidase: 6.5 ± 1.0 mM.

Optimum temperature: about 40-50 °C (at a pH of about 6-8)

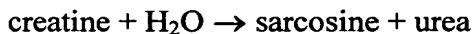
Optimum pH: pH about 8.0-9.0 (at a temperature of about 37° C)

Molecular weight: about 43,000 (SDS-PAGE).

*3
A
Cont*

37. (Amended) A creatine amidinohydrolase (i) encoded by a nucleic acid sequence obtained by mutation of (a) the nucleic acid sequence of SEQ ID NO:2 or (b) a nucleic acid sequence encoding the amino acid sequence of SEQ ID NO:1 and (ii) having the following physicochemical properties:

Action: catalyzing the following reaction:



Heat stability: not more than about 50 °C (pH 7.5, 30 min)

pH stability: being stable at pH 5-8 (40 °C, 18 h preservation).

K_m values for creatine in a coupling assay using a sarcosine oxidase and a peroxidase: 9.0 ± 1.0 mM.

Optimum temperature: about 40-50 °C (at a pH of about 6-8)

Optimum pH: pH about 8.0-9.0 (at a temperature of about 37° C)